

DETAILED ACTION

Applicant's arguments filed 7/28/08 have been fully considered but they are not persuasive. The amendment has been entered. Claims 1-6, 8-12 are pending. Claims 7, 13-18 are canceled.

Claims 1-6, 8-12 are under consideration.

Claim Objections

Claims 4, 5, 6, 8, 9, 11, 12 objection to under 37 CFR 1.75(c) as being in improper form because a multiple dependent claim should refer to other claims in the alternative only and cannot depend from any other multiple dependent claims is withdrawn in view of the amendment.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

(a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negated by the manner in which the invention was made.

Claims 1-5, 8-12 stand rejected under 35 U.S.C. 103(a) as being unpatentable over Nakagawa et al, (European Journal of Pharmaceutical Sciences, 13: 53-60, 2001) in view of Fitzsimons et al, (Gene Therapy, 8: 1675-1681, 2001).

Nakagawa teaches a recombinant adenovirus vector, which contains a transgene encoding for IL-12 controlled by the tetracycline-regulated expression system (p 55, 2nd column, p 56 1st and 2nd column). Nakagawa teaches the utility of both two-component and one-component systems tetracycline-regulatable adenovirus vectors. Two component-systems utilize one adenovirus vector to express the transgene under the control of the TRE and a minimal promoter, and a second adenovirus vector to express the transactivator, either tTA or rTA, from a constitutive promoter (p 55, 2nd column, p 56 1st column, last paragraph).

Furthermore, Nakagawa teaches a one component-system, wherein both expression cassettes are incorporated into a single adenovirus vector (p 55, 1st column, last paragraph and 2nd column, 1st paragraph and reference by incorporation). Nakagawa also teaches that, the tetracycline-sensitive one component system incorporated both expression cassettes into a single adenovirus vector, wherein the transgene is a nucleic acid sequence encoding the interleukin-12 transgene as claimed in the instant application, (claim 5). Nakagawa also teaches that, the tetracycline-sensitive one component system incorporate both expression cassettes into a single adenovirus vector, wherein the insert is inserted into the E1/E3-deleted backbone of Ad5 [reference by incorporation, (Corti et al, 1999), p 55, 2nd column, 1st sentence] as claimed in the instant application, (claim 8). Nakagawa also teaches that the tet-on system a "reverse" transactivator (rTA) with the opposite properties of tTA binds to the TRE and activates transcription only in the presence of tetracycline derivatives like doxocycline (p 54, 2nd column, last paragraph). Nakagawa teaches this tet adenovirus system provides new opportunities and improved safety for gene therapy applications in humans. Nakagawa teaches because systemic administration of IL-12 is toxic a regulated a temporal control and basal levels of the gene is essential (p 58). Nakagawa teaches by intratumoral injection of the vector in tumor bearing mice the hIL-12-mediated antitumor response was not compromised by reducing

intratumoral IL-12 concentrations during later stages of the therapy (p 58 columns 1-2).

Nakagawa differs from the present invention for not teaching the tet inducible cassette in the general structure as the claimed invention.

However, at the time of the instant invention Fitzsimons teaches a recombinant adeno-associated virus (rAAV) viral vector which contains an insert exhibiting the general structure in which, a) the TetO₇ is the heptamerized tetracycline operator; b) TK⁺ is the minimal thymidine kinase promoter; c) tTA is a nucleic acid sequence which encodes a fusion protein from the repressor protein inducible by tetracycline and the transcriptional activation domain of the Herpes simplex virus VP16, d) CMV is the minimal cytomegalovirus promoter; e) the transgene is a nucleic acid sequence which codes for a non-viral protein luciferase; f) intron¹ is a desired non-encoding nucleic acid sequence insulator with a length of 42 bp; and g) intron² is a desired non-encoding nucleic acid sequence insulator with a length of 42 bp (p 1675, 2nd column, last paragraph and p 1676, 1st column and figure 1) as is claimed in the instant case. Fitzsimons also teaches they have optimized the autoregulated-directional rAAV-based construct for in vitro and in vivo regulation of gene expression by doxycycline and they have demonstrated that rAAV-mediated transfer of reporter genes which can be regulated in vitro and in vivo with extremely low basal expression (p 1675, 1st column, 2nd paragraph). Fitzsimons also teaches they have minimized the size of the cassette and decreased the basal leakiness of the system, leading to tight regulation in the rat brain (abstract). Fitzsimons suggests the ideal regulatory system would be one in which all components were contained in one vector genome thus requiring a cell to be transduced with one vector rather than two or three (p 1679, 1st column, 2nd paragraph). Fitzsimons teaches that the bidirectional system was found to be appropriately expressed and regulated and used to transduce HEK 293 cells in vitro and the insulators may act more effectively in vivo to limit interactions between the ITRs and the expression cassette (p

1679, 2nd column). As such Fitzsimons provide sufficient motivation to apply the bidirectional tet-inducible cassette into the adenovirus of Nakagawa to tightly regulate IL-12 levels for gene therapy.

Accordingly, in view of the teachings of Fitzsimons et al, it would have been obvious for one of ordinary skill in the art, at the time the claimed invention was made, to modify the adenovirus vector of Nakagawa and insert the tetracycline-regulated cassette of Fitzsimons into the adenovirus for gene therapy with a reasonable expectation of success. One of ordinary skill in the art would have been sufficiently motivated to make such a modification since Nakagawa teaches the temporal control and the basal levels of exogenous gene expression is essential for IL-12 gene therapy and teaches efficacy and safety of tetracycline-regulated adenovirus-mediated IL-12 gene therapy for prostate cancer (p 58). Moreover, Nakagawa suggests that adenovirus vectors with tetracycline-regulated gene expression may prove useful not only for cytokine gene therapy applications, but also in other gene therapy applications, such as those of neurodegenerative diseases where, temporal control of exogenous gene expression is essential (p 58 under conclusion). One of ordinary skill in the art would have been particularly motivated to introduce the tet-regulated cassette of Fitzsimons into an adenovirus since Fitzsimons suggests the ideal regulatory system would be one in which all components were contained in one vector genome thus requiring a cell to be transduced with one vector rather than two or three and that the bidirectional system was found to be appropriately expressed and regulated in cells *in vitro* and the insulators may act more effectively *in vivo* to limit interactions between the ITRs and the expression cassette of the bidirectional tet-inducible cassette will tightly regulate IL-12 levels for gene therapy.

Thus, the claimed invention as a whole, is clearly *prima facie* obvious in the absence of evidence to the contrary.

A. Applicant argue: (1) The references, viewed alone or in combination, do not teach or suggest all of the elements of the rejected claims, and (2) the claimed invention represents more than the predictable use of the elements described in the cited prior art as evidenced by the unexpected results described in detail below. As disclosed in the specification, the claimed invention is more than the predictable use of prior art elements according to their established functions. The current Office Action misconstrues the concept of what constitutes unexpected results. In particular, the Office argues that unexpected results are precluded because the Fitzsimons vector has some of the same elements as the claimed vector (page 8, final sentence). This statement conveys a lack of understanding of the doctrine of unexpected results, which necessarily always applies in situations where each cited prior art reference contains at least some of the same elements as the claimed invention. Without prior art claiming elements of the invention, there would be no need to point out the unexpected results.

These arguments are not persuasive because the office action on page 8, final sentence states "... the tet inducible construct of Fitzsimons has the same structure as the tet inducible cassette construct of the claimed invention (see Fitzsimons, p 1676, figure 1, construct – BiG3.5)". This statement conveys the structure of the claimed product is the same as the structure product of the art, therefore it conveys any unexpected results of the art are obvious over any unexpected results of the claimed instant invention and does not misconstrue the concept of what constitutes unexpected results whatsoever. Applicant's arguments are not persuasive because in the instant case Fitzsimons taken with Nakagawa provide all the structural elements of the claimed vector as required in the instant invention.

B. Applicants the question is whether the result is synergistic and adds up to more than the sum of its parts, more what would be expected by the skilled person. In their previous

response, Applicants pointed to the following evidence of unexpected results: A) Expression of the construct described by Fitzsimons et al. was less than two-fold higher as compared to expression in a plasmid (see Figure 4). Suppression (repression) is shown between 100-fold to 200-fold with a maximum at 204-fold (Block et al., Journal of Gene Medicine 5:190-200 (2003), Exhibit 1 to Applicants submission filed November 5, 2007, Figures 5 and 7). Fitzsimons only discloses constructs expressing luciferase, whereas constructs containing IL12 or other transgenes are not disclosed at all.

These arguments are not persuasive because a) the construct provided by Fitzsimons is taken with the construct provided by Nakagawa and the post filing evidence of comparison provided in the form of arguments by the Applicant is not obvious for said comparison; b) the tet inducible cassette construct of Fitzsimons has the same structure as the tet inducible cassette construct of the claimed invention (see Fitzsimons, p 1676, figure 1, construct pBiG3r.5). The difference between the Fitzsimons, vector and the claimed vector is that the Fitzsimons, vector is a rAAV vs the claimed adenovirus vector. However, upon the insertion of the Fitzsimons tet inducible cassette into the adenovirus cassette of Nakagawa inherently the end product of the combined cited references will produce the unexpected results of the claimed invention. The comparison between the Fitzsimons system and the vector of the invention does not provide lack of *prima facie* because lacks comparison between the vector of the combined cited references and the vector of the invention. The art of introducing DNA cassettes from a rAAV into an adenovirus is high and an ordinary artisan in the art of IL-12 gene therapy would have been sufficiently motivated to replace the rAAV with an adenovirus of the combined cited references. Fitzsimons and Nakagawa provides motivation to insert the claimed tet inducible cassette into an adenovirus by suggesting the tet adenovirus system provides new opportunities and improved safety for gene therapy. Nakagawa particularly provides motivation by teaching

an immunotherapy model for prostate cancer with a tetracycline-regulated adenovirus vector expressing the cytokine IL-12. Moreover, Nakagawa teaches the efficacy of the tetracycline-regulated IL-12 gene expression in RM-1 cells and suggests that this adenovirus-based tTA tet system may be useful for regulated gene expression in broad range of mammalian cells and to create effective IL-12 vaccine and the adenovirus vector with tetracycline-regulated gene expression may prove useful not only for gene therapy of cancer but also for gene therapy applications such as those of neurodegenerative diseases where the temporal control of exogenous gene expression is essential; c) Comparison With Closest Prior Art An affidavit or declaration under 37 CFR 1.132 must compare the claimed subject matter with the closest prior art to be effective to rebut a *prima facie* case of obviousness. *In re Burckel*, 592 F.2d 1175, 201 USPQ 67 (CCPA 1979). "A comparison of the claimed invention with the disclosure of each cited reference to determine the number of claim limitations in common with each reference, bearing in mind the relative importance of particular limitations, will usually yield the closest single prior art reference." *In re Merchant*, 575 F.2d 865, 868, 197 USPQ 785, 787 (CCPA 1978) (emphasis in original). Where the comparison is not identical with the reference disclosure, deviations therefrom should be explained, *In re Finley*, 174 F.2d 130, 81 USPQ 383 (CCPA 1949), and if not explained should be noted and evaluated, and if significant, explanation should be required. *In re Armstrong*, 280 F.2d 132, 126 USPQ 281 (CCPA 1960) (deviations from example were inconsequential). See 716.02(e).

C) Applicants argue gene expression for the claimed adenoviral vector construct is much higher than for the Fitzsimons et al. rAAV vector. For example, for a construct containing IL12 as transgene, up to 1,000 pg/24 h/106 cells could be expressed in a linear, m.o.i., dependent

fashion for various cell lines using the same transgene (IL12) in 48 hours at a m.o.i. of 10 (expression of up to 200 pg). (Block et al., Journal of Gene Medicine 5:190-200 (2003)).

Applicants failed to provide evidence that for example, the post filing construct containing IL12 as

transgene of Block et al., Journal of Gene Medicine 5:190-200 (2003)) would provide gene expression much higher than for example the adenoviral tet IL-12 vector of Fitzsimons/Nakagawa. Comparison With Closest Prior Art An affidavit or declaration under 37 CFR 1.132 must compare the claimed subject matter with the closest prior art to be effective to rebut a *prima facie* case of obviousness. In re Burckel, 592 F.2d 1175, 201 USPQ 67 (CCPA 1979). "A comparison of the claimed invention with the disclosure of each cited reference to determine the number of claim limitations in common with each reference, bearing in mind the relative importance of particular limitations, will usually yield the closest single prior art reference." In re Merchant, 575 F.2d 865, 868, 197 USPQ 785, 787 (CCPA 1978) (emphasis in original). Where the comparison is not identical with the reference disclosure, deviations therefrom should be explained, In re Finley, 174 F.2d 130, 81 USPQ 383 (CCPA 1949), and if not explained should be noted and evaluated, and if significant, explanation should be required. In re Armstrong, 280 F.2d 132, 126 USPQ 281 (CCPA 1960) (deviations from example were inconsequential). See 716.02(e).

D) Applicants argue high efficiency of gene expression was confirmed in various cell lines and for various transgenes with a comparable order of magnitude and 6,000-fold for IL12 expression. The order of magnitude of suppression was also completely surprising and unexpected by the skilled artisan. Exhibit 2 to Applicants submission filed November 5, 2007 The claimed construct provides 16,000-fold suppression for expression of the luciferase gene, is Figure 1, which shows expression of hIL12 following infection of human colon carcinoma cells

with 10 m.o.i, and incubation over 24 hours using various concentrations of doxycyclin using the claimed adenoviral vectors. Determination of ILI2 in supernatant and cell lysate is shown. The data obtained for the claimed vectors according to claim 1 support the unexpected high native expression and suppressibility achieved with the claimed vectors and represent their application in clinical studies.

These arguments are not persuasive because regarding the vector of the invention provides 6000-fold Dox-regulated suppressibility (IL-12) vs not tested of Fitzsimons et al does not provide unexpected results because where the unexpected properties of a claimed invention are not shown to have a significance equal to or greater than the expected properties, the evidence of unexpected properties may not be sufficient to rebut the evidence of obviousness. See MPEP 716.02(c). Regarding the claimed construct provides 16,000-fold suppression for expression of the luciferase gene Applicants failed to provide evidence for example of comparison of the vector of the invention as compared to for example the adenoviral tet IL-12 vector of Fitzsimons/Nakagawa Dox-regulated suppressibility (Luc) to overcome the art rejection. Regarding exhibit 2 lacks figure 1 which shows expression of hIL2 following infection of human colon carcinoma cells, therefore no response is applicable. Again, attorney arguemnts cannot take place of evidence in the record. In re Schulze, 346 F.2d 600, 602, 145 USPQ 716, 718 (CCPA 1965). Examples of attorney statements which are not evidence and which must be supported by an appropriate affidavit or declaration include statements regarding unexpected results, commercial success, solution of a long-felt need, inoperability of the prior art, invention before the date of the reference, and allegations that the author(s) of the prior art derived the disclosed subject matter from the applicant. See MPEP § 2145 generally for case law pertinent to the consideration of applicant's rebuttal arguments.

E) Applicants argue due to the unexpected high suppressibility of ILI2 expression, complete protection in C57B 16 mice could be achieved by adding doxycyclin (Dox) to the drinking water following systemic application of a 100 % lethal vector dosage. Further, attached as Exhibit 3 to Applicants submission filed November 5, 2007, is Figure 2, which shows that intra-tumoral application led to highly significant tumor regression in all treated animals showing a long-term survival of 25%. Attached as Exhibit 4 to Applicants submission filed November 5, 2007, is Figure 3, which shows tumors after day 18. Well observable are large tumors under control (mock) or when using control vector Ad.DL312, whereas no tumors or only very small tumors were observed following application of vector Ad3r-IL12 of the invention. Applicants argue For the Examiner's convenience, the significant differences of the vector according to the invention as compared to the expression system according to Fitzsimons were shown in Table I of Applicants' submission filed November 5, 2007.

These arguments are not persuasive because attorney arguments cannot take place of evidence in the record of inoperability of the prior art. *In re Schulze*, 346 F.2d 600, 602, 145 USPQ 716, 718 (CCPA 1965). Examples of attorney statements which are not evidence and which must be supported by an appropriate affidavit or declaration include statements regarding unexpected results, commercial success, solution of a long-felt need, inoperability of the prior art, invention before the date of the reference, and allegations that the author(s) of the prior art derived the disclosed subject matter from the applicant. See MPEP 716.01

Claim 6 stands rejected under 35 U.S.C. 103(a) as being unpatentable over **Nakagawa et al.** (*European Journal of Pharmaceutical Sciences*, 13: 53-60, 2001) in view of **Fitzsimons et**

al, (Gene Therapy, 8: 1675-1681, 2001) as applied to claims 1-5, 8-12 above, and further in view of **Lode et al.** (European Journal of Pharmaceutical Sciences, 13: 53-60, 2001).

The 103 rejection of claims 1-5, 8-12 as being unpatentable over Nakagawa taken with Fitzsimons is applied here as indicated above.

Nakagawa taken with Fitzsimons do not teach IL-12 is a single chain interleukin-12.

However, at the time the invention was made, Lode is an exemplified prior art that teaches that it is routine or well-established in the art to employ a single chain IL-12 for protective immunity. Lode et al, teaches a single chain IL-12 fusion protein induces T cell dependent protective immunity in a syngeneic model of murine neuroblastoma. Lode teaches the single chain IL-12 fusion protein induces a T cell mediated immunity that completely protects mice from challenge with the wild type tumor cells as indicated by the complete absence of liver and bone marrow metastases in a novel syngeneic model of neuroblastoma (p 2475, 2nd column). Lode teaches the poor immunogenicity of this model clearly demonstrates the feasibility of efficient gene therapy with a single chain IL-12 fusion protein.

Thus, it would also have been obvious for one of ordinary skill in the art of IL-12 immunoengic composition to employ the single chain IL-12 of choice available in the art in the immunogenic composition of IL-12 of the combined cited references. One of ordinary skill in the art would have been motivated to employ the single chain IL-12 for gene therapy in order to demonstrate successful anti-tumor immunotherapy with a single chain IL-12 fusion protein that would facilitate clinical application of IL-12 gene therapy as suggested by Lode et al and particularly in view of the totality of the prior art at the time the invention was made. One of ordinary skill in the art would have been motivated to employ the single chain IL-12 since Lode et al have demonstrated that subcutaneous vaccination with IL-12 single chain fusion protein induces a T cell-mediated immunity that completely protects mice from challenge with wild type

tumor cells as indicated by the complete absence of liver and bone marrow metastasis in a syngeneic model of neuroblastoma (p 2475, 2nd column. 2nd paragraph).

Thus, the claimed invention as a whole, is clearly *prima facie* obvious in the absence of evidence to the contrary.

Applicants argue the unexpected results achieved with the presently claimed vectors are described in the previously submitted exhibits and accompanying remarks submitted November 5, 2007, and reiterated above with additional arguments. Applicants maintain that the secondary reference by Lode et al. does not add anything of significance except that the authors teach single-chain IL12 fusion protein. The publication is completely silent as to the cassette bearing the transgene and the combination of Fitzsimons et al. and Lode et al. provides no expectation of the results described above.

In view of the arguments above and the previously submitted exhibits and accompanying remarks submitted November 5, 2007.

These arguments are not persuasive for the same reasons as discussed above regarding the unexpected results achieved by the claimed vectors. Regarding Fitzsimons and Nakagawa provide motivation for an adenovirus tet inducible system as the claimed invention and since Lode teaches single chain IL-12 fusion protein induces T cell dependent protective immunity in a syngeneic murine neuroblastoma model and suggests the feasibility of efficient gene therapy with a single IL-12 fusion gene therefore, Lode et al. provide sufficient motivation for introducing the single chain IL-12 in a tet inducible system of an adenovirus containing the bicistronic cassette of Fitzsimons.

Conclusion

No claim is allowed.

THIS ACTION IS MADE FINAL. Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Magdalene K. Sgagias whose telephone number is (571) 272-3305. The examiner can normally be reached on Monday through Friday from 9:00 am to 5:00 pm. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Peter Paras, Jr., can be reached on (571) 272-4517. The fax phone number for the organization where this application or proceeding is assigned is (703) 872-9306.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should you have questions on access to private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll free).

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